Mitochondrial Diversity of Human Head Lice (*Pediculus humanus capitis*) Across the Americas

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Background

•Sucking lice (Phthiraptera: Anoplura) are permanent and obligate ectoparasites of eutherian mammals. These highly specialized blood-sucking insects live in close association with their hosts where they complete their entire life cycle. This relationship has led to coevolution in which parasite diversification parallels that of the host (Figure 3).

• The human head louse has coevolved with humans over millions of years (Figure 1, Figure 2). Therefore, louse molecular data is valuable for studying the evolutionary history of lice and their human hosts.

•Human head lice are genetically diverse and have 3 deeply divergent mitochondrial (mtDNA) clades, named A, B, and C.

•Clothing lice (clade A) has been known to carry three bacterial pathogens. Genotypic louse data would be valuable to determine which clades of head lice have the potential to be carriers in order to combat resistance to pediculcides.

•Previous studies suggest that the three head louse clades have different evolutionary histories. Clade A appears to be uniquely linked to its *Homo sapiens* host (modern humans). Clade B, however, appears to have evolved on an archaic hominin in Europe (possibly *H. neanderthalensis*) and later switched to modern *H. sapiens*. Clade C appears to have evolved on an archaic hominin in Asia (possibly *H. erectus*) or Africa.

Figure 1: Male head louse

Figure 2: Early lice comb

Evolutionary History of Lice

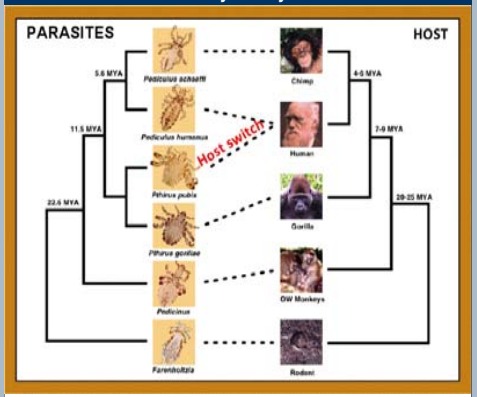

Figure 3: The coevolutionary history of lice and Great Apes. Humans are parasitized by the head and clothing louse (*Pediculus humanus*), and the crab louse (*Pthirus pubis*).

Methods

•In this study, we used the COX1 gene to analyze mtDNA from 450 lice in the Americas to better understand human colonization of the New World.

•The forward and reverse sequences were aligned using Sequencher 4.5 (Gene Codes Corporation). Unique haplotypes were determined using using DnaSP v. 4.10.9 software (Rozas et al. 2003). A matrix of pairwise differences was calculated with MEGA version 4 (Tamura et al. 2007). The genetic relationships were estimated by constructing neighbor-joining (Saitou and Nei 1987) trees using PAUP* 4.0b10 (Wilgenbusch and Swofford 2003).

•Two outgroup COX1 sequences from chimp louse (*Pediculus schaeffi*) were aligned to the head louse dataset .The genealogical relationships among haplotypes within each human louse mitochondrial clade were analyzed using the program TCS version 1.13 (Clement, Posada, and Crandall 2000).

•Haplotype frequencies for the Americas were determined and graphed using Microsoft Excel.

Results

•Among the 450 sequences, we found 12 haplotypes belonging to clade A (N=333) and 25 in clade B (N=117).

• Haplotype frequencies differ geographically. Haplotypes A and B were both relatively common in North America, with 58% haplotype A and 42% haplotype B. In Central America, haplotype A was much less common than haplotype B (18% and 82%, respectively). South America consisted of 95% haplotype A and only 5% haplotype B.

•Sequence comparisons for alignment of 379 nucleotides revealed 47 substitutions.

Phylogenetic Tree

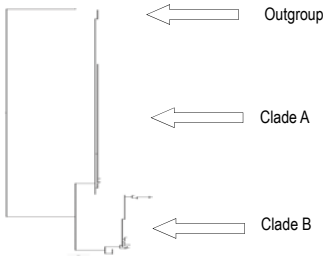

Figure 4: Although specimen localities are not shown in the figure due to space constraints, there are almost no clades that are exclusively from one geographic area, which tells us that there is no obvious phylogeographical structure in this tree.

Statistical Parsimony Networks

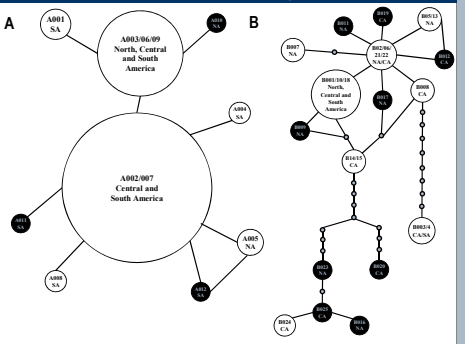

Figure 5: Statistical parsimony networks for the COX1 haplotypes found in clade A (A) and in clade B (B). Each connecting branch represents a single mutational step and inferred missing intermediate haplotypes are represented by open circles. Size are scaled and represent relative frequencies, see table 2 for absolute frequencies. Circles with black background and white letters are singletons (unique haplotypes found in a single location). For each haplotype the geographic distribution as North (NA), Central (CA) and South America (SA) is indicated. Mexico was included with the Central America countries due to the different colonization history than the rest of North America.

Haplotype Distribution

Table 1. Sampling sites and geographic distribution of mitochondrial DNA haplotypes based on mitochondrial sequences from the COI gene (379 bp) in human head lice (*Pediculus humanus capitis*) from the Americas.

			Haplogroup	
			A	**B**
North America	USA	California	13	0
		Utah	13	15
		Washington	51	8
		Florida	8	15
		Georgia	0	2
		New York	9	5
		Tennessee	0	4
		Texas	2	20
	Mexico		4	10
	Subtotal		100	79
Central America		Honduras	4	25
		Panama	0	2
	Subtotal		4	27
South America		Argentina	224	11
		Colombia	2	0
		Ecuador	2	0
		Peru	1	0
	Subtotal		229	11
TOTAL	N=450		333	117

Distribution Map

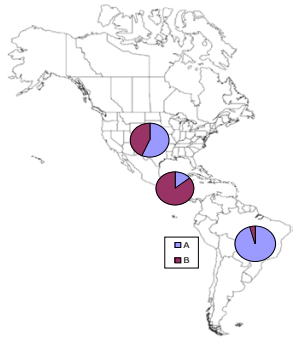

Figure 6: We used haplotype frequencies to describe the proportion of mitochondrial haplotypes belonging to clade A (blue) and B (purple) in each of the Americas: North, Central and South.

Discussion

•We suggest different human colonization patterns in the New World could account for the differing haplotype frequencies seen today in the human louse parasite populations.

•The low number of A-haplotypes supports the idea that clade A might have evolved in modern humans because they show the same genetic signature of a bottleneck during the out-of-Africa expansion.

•Clade B might have coevolved with Neanderthals which were distributed mostly in Europe. Because of the European colonization of the New World, it is possible that some of the haplotype B lice were derived from louse populations brought by Europeans.

•Considering the ancient distribution of head lice throughout the Americas and our mitochondrial data, we hypothesize that the first people of the Americas carried lice from both haplogroups A and B.

• Genetic analysis of both the causative agent of the disease and the lice will help in understanding the origin of outbreaks and disease dynamics, and in the developing of control strategies.

•More markers including highly nuclear polymorphic loci, such as microsatellites, are needed to further analyze the genetic diversity of human lice in the Americas.

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